

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

**Listing of the Claims:**

1. (Original) A conjugate of an affinity molecule and a water-dispersible nanoparticle, wherein the nanoparticle comprises (a) an inner core comprised of a semiconductive or metallic material, (b) a water-insoluble organic coating provided thereon, (c) an outer layer of a multiply amphipathic polymer having at least two hydrophobic regions and at least two hydrophilic regions, and (d) a functional group directly or indirectly linked to the multiply amphipathic polymer, and further wherein the affinity molecule is bound to the functional group.
2. (Original) The conjugate of claim 1, wherein the functional group is directly linked to the multiply amphipathic polymer.
3. (Original) The conjugate of claim 1, wherein the functional group is indirectly linked to the multiply amphipathic polymer through an inert linking moiety.
4. (Original) The conjugate of claim 1, wherein the affinity molecule is selected so as to specifically bind to a biological target molecule.
5. (Original) The conjugate of claim 4, wherein the affinity molecule specifically binds to the biological target molecule through noncovalent interaction.
6. (Original) The conjugate of claim 4, wherein the affinity molecule specifically binds to the biological target molecule through covalent interaction.
7. (Original) The conjugate of claim 4, wherein the affinity molecule is selected from the group consisting of an antigen, hapten, antibody, antibody fragment, hormone, hormone binding protein, enzyme, enzyme inhibitor, nucleic acid, nucleotide, oligonucleotide, polynucleotide, receptor agonist, and receptor antagonist.
8. (Original) The conjugate of claim 1, wherein the functional group is selected from amino, cyano, carboxyl, ester, and halocarbonyl groups.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

9. (Original) The conjugate of claim 8, wherein the functional group is an amino group.

10. (Previously presented) The conjugate of claim 1, wherein the inner core of the nanoparticle is comprised of a semiconductive material.

11. (Original) The conjugate of claim 10, wherein the semiconductive material is inorganic.

12. (Original) The conjugate of claim 11, wherein the semiconductive material is crystalline.

13. (Original) The conjugate of claim 10, wherein the water-insoluble organic coating is comprised of trioctylphosphine oxide, trioctylphosphine, tributylphosphine, or a mixture thereof.

14. (Original) The conjugate of claim 10, further including a shell layer between the core and the water-insoluble organic coating.

15. (Original) The conjugate of claim 14, wherein the shell layer is comprised of a semiconductive material having a band gap energy greater than that of the inner core.

16. (Original) The conjugate of claim 1, wherein the inner core is comprised of a metallic material.

17. (Original) The conjugate of claim 16, wherein the water-insoluble organic coating has an affinity for the metallic material.

18. (Original) The conjugate of claim 12, wherein the water-insoluble organic coating is comprised of a hydrophobic surfactant.

19. (Previously presented) The conjugate of claim 18, wherein the hydrophobic surfactant is selected from the group consisting of octanethiol, dodecanethiol, dodecylamine, tetraoctylammonium bromide, and mixtures thereof.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

20. (Original) The conjugate of claim 1, wherein the multiply amphipathic polymer is linear or branched.
21. (Original) The conjugate of claim 20, wherein the multiply amphipathic polymer is branched.
22. (Original) The conjugate of claim 21, wherein the multiply amphipathic polymer is hyperbranched or dendritic.
23. (Original) The conjugate of claim 1, wherein the hydrophobic regions of the multiply amphipathic polymer are each comprised of at least one non-ionizable, nonpolar monomer unit.
24. (Original) The conjugate of claim 1, wherein each of the hydrophobic regions comprise at least one monomer unit selected from the group consisting of ethylene, propylene, alkyl (C<sub>4</sub>-C<sub>12</sub>)-substituted ethyleneimine, an alkyl acrylate, a methacrylate, a phenyl acrylate, an alkyl acrylamide, a styrene, a hydrophobically derivatized styrene, a vinyl ether, a vinyl ester, a vinyl halide, and a combination thereof.
25. (Original) The conjugate of claim 24, wherein the hydrophobic regions comprise at least one monomer unit selected from an alkyl acrylate, an alkyl methacrylate, an alkyl acrylamide, and a mixture thereof.
26. (Original) The conjugate of claim 1, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit containing an ionizable or polar moiety.
27. (Original) The conjugate of claim 26, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.
28. (Original) The conjugate of claim 27, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and amine substituents.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

29. (Original) The conjugate of claim 23, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable or polar moiety.

30. (Original) The conjugate of claim 29, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

31. (Original) The conjugate of claim 30, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and an amine substituent.

32. (Original) The conjugate of claim 1, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit selected from the group consisting of a water-soluble ethylenically unsaturated C<sub>3</sub>-C<sub>6</sub> carboxylic acid, an allylamine, an inorganic acid addition salt of an allylamine, a di-C<sub>1</sub>-C<sub>3</sub>-alkylamino-C<sub>2</sub>-C<sub>6</sub>-alkyl acrylate, a methacrylate, an olefinically unsaturated nitrile, a diolefinically unsaturated monomer, N-vinyl pyrrolidone, N-vinyl formamide, an acrylamide, a lower alkyl-substituted acrylamide, a lower alkoxy-substituted acrylamide, N-vinylimidazole, N-vinylimidazoline, a styrene sulfonic acid and an alkylene oxide.

33. (Original) The conjugate of claim 32, wherein the hydrophilic regions each comprise at least one monomer unit selected from the group consisting of acrylic acid, methacrylic acid, styrene sulfonic acid, acrylamide and methacrylamide.

34. (Original) The conjugate of claim 1, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise a vinyl monomer substituted with at least one hydrophilic moiety selected from the group consisting of a carboxylate, a thiocarboxylate, an amide, an imide, a hydrazine, a sulfonate, a sulfoxide, a sulfone, a sulfite, a phosphate, a phosphonate, a phosphonium, an alcohol, a thiol, a nitrate, an amine, an ammonium, and an alkyl ammonium group  $[\text{NHR}^1\text{R}^2]^+$ , wherein R<sup>1</sup> and R<sup>2</sup> are alkyl substituents.

35. (Original) The conjugate of claim 34, wherein the hydrophilic moiety is directly bound to a carbon atom in the backbone of the multiply amphipathic polymer.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

36. (Original) The conjugate of claim 34, wherein the hydrophilic moiety is bound to a carbon atom in the polymer backbone of the multiply amphipathic polymer through a linkage selected from the group consisting of alkylene, alkenylene, heteroalkylene, heteroalkenylene, arylene, heteroarylene, alkarylene, and aralkylene.

37. (Original) The conjugate of claim 1, wherein the multiply amphipathic polymer is a copolymer of a hydrophilic monomer selected from the group consisting of acrylic acid, methacrylic acid and combinations thereof, with at least one hydrophobic alkyl (C<sub>6</sub>-C<sub>12</sub>) acrylamide monomer.

38. (Original) The conjugate of claim 37, wherein the multiply amphipathic polymer is poly(acrylic acid-co-octylacrylamide).

39. (Original) The conjugate of claim 1, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 500 to 50,000.

40. (Original) The conjugate of claim 39, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 10,000.

41. (Original) The conjugate of claim 40, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 5000.

42. (Original) The conjugate of claim 38, wherein the poly(acrylic acid-co-octylacrylamide) has a molecular weight in the range of approximately 1000 to 5000.

43. (Original) The conjugate of claim 1, wherein the hydrophobic regions represent in the range of approximately 25 wt. % to 90 wt. % of the multiply amphipathic polymer.

44. (Original) The conjugate of claim 1, wherein the multiply amphipathic polymer is a polypeptide in which the hydrophobic regions are comprised of at least one hydrophobic amino acid and the hydrophilic regions are comprised of at least one hydrophilic amino acid.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

45. (Original) The conjugate of claim 1, wherein the multiply amphipathic polymer is crosslinked.

46. (Original) A composition comprising a plurality of nanoparticle conjugates each composed of a water-dispersible nanoparticle and an affinity molecule, wherein each water-dispersible nanoparticle comprises (a) an inner core comprised of a semiconductive or metallic material, (b) a water-insoluble organic coating provided thereon, and (c) an outer layer of a multiply amphipathic polymer having at least two hydrophobic regions and at least two hydrophilic regions, and (d) a functional group directly or indirectly linked to the polymer, and further wherein the affinity molecule is bound to the functional group.

47. (Original) The composition of claim 46, wherein the functional group is directly linked to the multiply amphipathic polymer.

48. (Original) The composition of claim 46, wherein the functional group is indirectly linked to the multiply amphipathic polymer through an inert linking moiety.

49. (Original) The composition of claim 46, wherein the inner cores of the nanoparticles are members of a monodisperse particle population.

50. (Original) The composition of claim 49, wherein the monodisperse particle population is characterized in that when irradiated the population emits light in a bandwidth in the range of approximately 20 nm to 60 nm full width at half maximum (FWHM).

51. (Original) The composition of claim 50, wherein the monodisperse particle population is characterized in that when irradiated the population emits light in a bandwidth in the range of approximately 20 nm to 40 nm full width at half maximum (FWHM).

52. (Original) The composition of claim 49, wherein the monodisperse particle population is characterized in that it exhibits no more than about a 10% rms deviation in the diameter of the inner core.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

53. (Original) The composition of claim 52, wherein the monodisperse particle population is characterized in that it exhibits no more than about a 5% rms deviation in the diameter of the inner core.

54. (Original) The composition of claim 46, wherein the affinity molecule is selected so as to specifically bind to a biological target molecule.

55. (Previously presented) The composition of claim 54, wherein the selected from the group consisting of an antigen, hapten, antibody, antibody fragment, hormone, hormone binding protein, enzyme, enzyme inhibitor, nucleic acid, nucleotide, oligonucleotide, polynucleotide, receptor agonist, and receptor antagonist.

56. (Original) The composition of claim 54, wherein the affinity molecule specifically binds to the biological target molecule through noncovalent interaction.

57. (Previously presented) The composition of claim 54, wherein the affinity molecule specifically binds to the biological target molecule through covalent interaction.

58. (Original) The composition of claim 46, wherein the functional group is selected from amino, cyano, carboxyl, ester, and halocarbonyl groups.

59. (Original) The composition of claim 58, wherein the functional group is an amino group.

60. (Original) The composition of claim 59, wherein the inner core is comprised of a semiconductive material.

61. (Original) The composition of claim 60, wherein the semiconductive material is inorganic.

62. (Original) The composition of claim 61, wherein the semiconductive material is crystalline.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

63. (Original) The composition of claim 60, wherein the water-insoluble organic coating is comprised of trioctylphosphine oxide, trioctylphosphine, tributylphosphine, or a mixture thereof.

64. (Original) The composition of claim 60, further including a shell layer between the core and the water-insoluble organic coating.

65. (Original) The composition of claim 64, wherein the shell layer is comprised of a semiconductive material having a band gap energy greater than that of the inner core.

66. (Previously amended) The composition of claim 59, wherein the inner core is comprised of a metallic material.

67. (Original) The composition of claim 66, wherein the water-insoluble organic coating has an affinity for the metallic material.

68. (Original) The composition of claim 67, wherein the water-insoluble organic coating is comprised of a hydrophobic surfactant.

69. (Original) The composition of claim 68, wherein the hydrophobic surfactant is selected from the group consisting of octanethiol, dodecanethiol, dodecylamine, tetraoctylammonium bromide, and mixtures thereof.

70. (Original) The composition of claim 46, wherein the multiply amphipathic polymer is linear or branched.

71. (Original) The composition of claim 70, wherein the multiply amphipathic polymer is branched.

72. (Original) The composition of claim 71, wherein the multiply amphipathic polymer is hyperbranched or dendritic.

73. (Original) The composition of claim 46, wherein the hydrophobic regions of the multiply amphipathic polymer are each comprised of at least one non-ionizable, nonpolar monomer unit.



Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

74. (Original) The composition of claim 46, wherein each of the hydrophobic regions comprise at least one monomer unit selected from the group consisting of ethylene, propylene, alkyl (C<sub>4</sub>-C<sub>12</sub>)-substituted ethyleneimine, an alkyl acrylate, a methacrylate, a phenyl acrylate, an alkyl acrylamide, a styrene, a hydrophobically derivatized styrene, a vinyl ether, a vinyl ester, a vinyl halide, and a combination thereof.

75. (Original) The composition of claim 74, wherein the hydrophobic regions comprise at least one monomer unit selected from an alkyl acrylate, an alkyl methacrylate, an alkyl acrylamide, and a mixture thereof.

76. (Original) The composition of claim 46, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit containing an ionizable or polar moiety.

77. (Original) The composition of claim 76, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

78. (Original) The composition of claim 77, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and amine substituents.

79. (Original) The composition of claim 73, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable or polar moiety.

80. (Original) The composition of claim 79, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

81. (Original) The composition of claim 80, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and an amine substituent.

82. (Original) The composition of claim 46, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit selected from the group consisting of a water-soluble ethylenically unsaturated C<sub>3</sub>-C<sub>6</sub> carboxylic acid, an

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

allylamine, an inorganic acid addition salt of an allylamine, a di-C<sub>1</sub>-C<sub>3</sub>-alkylamino-C<sub>2</sub>-C<sub>6</sub>-alkyl acrylate, a methacrylate, an olefinically unsaturated nitrile, a diolefinically unsaturated monomer, N-vinyl pyrrolidone, N-vinyl formamide, an acrylamide, a lower alkyl-substituted acrylamide, a lower alkoxy-substituted acrylamide, N-vinylimidazole, N-vinylimidazoline, a styrene sulfonic acid and an alkylene oxide.

83. (Original) The composition of claim 82, wherein the hydrophilic regions each comprise at least one monomer unit selected from the group consisting of acrylic acid, methacrylic acid, styrene sulfonic acid, acrylamide and methacrylamide.

84. (Original) The composition of claim 46, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise a vinyl monomer substituted with at least one hydrophilic moiety selected from the group consisting of a carboxylate, a thiocarboxylate, an amide, an imide, a hydrazine, a sulfonate, a sulfoxide, a sulfone, a sulfite, a phosphate, a phosphonate, a phosphonium, an alcohol, a thiol, a nitrate, an amine, an ammonium, and an alkyl ammonium group  $-\text{[NHR}^1\text{R}^2\text{]}^+$ , wherein R<sup>1</sup> and R<sup>2</sup> are alkyl substituents.

85. (Original) The composition of claim 84, wherein the hydrophilic moiety is directly bound to a carbon atom in the backbone of the multiply amphipathic polymer.

86. (Original) The composition of claim 84, wherein the hydrophilic moiety is bound to a carbon atom in the polymer of the multiply amphipathic polymer through a linkage selected from the group consisting of alkylene, alkenylene, heteroalkylene, heteroalkenylene, arylene, heteroarylene, alkarylene, and aralkylene.

87. (Original) The composition of claim 46, wherein the multiply amphipathic polymer is a copolymer of a hydrophilic monomer selected from the group consisting of acrylic acid, methacrylic acid and combinations thereof, with at least one hydrophobic alkyl (C<sub>6</sub>-C<sub>12</sub>) acrylamide monomer.

88. (Original) The composition of claim 87, wherein the multiply amphipathic polymer is poly(acrylic acid-co-octylacrylamide).

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

89. (Original) The composition of claim 46, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 500 to 50,000.

90. (Original) The composition of claim 89, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 10,000.

91. (Original) The composition of claim 90, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 5000.

92. (Original) The composition of claim 88, wherein the poly(acrylic acid-co-octylacrylamide) has a molecular weight in the range of approximately 1000 to 5000.

93. (Original) The composition of claim 46, wherein the hydrophobic regions represent in the range of approximately 25 wt. % to 90 wt. % of the multiply amphipathic polymer.

94. (Original) The composition of claim 46, wherein the multiply amphipathic polymer is a polypeptide in which the hydrophobic regions are comprised of at least one hydrophobic amino acid and the hydrophilic regions are comprised of at least one hydrophilic amino acid.

95. (Original) The composition of claim 46, wherein the multiply amphipathic polymer is crosslinked.

96. (Original) A nanoparticle conjugate comprising:

(a) a water-dispersible nanoparticle and an affinity molecule that serves as a first member of a binding pair, wherein the nanoparticle comprises (i) an inner core comprised of a semiconductive or metallic material, (ii) a water-insoluble organic coating provided thereon, (iii) an outer layer of a multiply amphipathic polymer having at least two hydrophobic regions and at least two hydrophilic regions, and (iv) a functional group covalently linked to the polymer, and further wherein the affinity molecule is bound to the functional group; and

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

(b) a second member of the binding pair associated with the first member through either covalent or noncovalent interaction.

97. (Original) The conjugate of claim 96, wherein the functional group is directly linked to the multiply amphipathic polymer.

98. (Original) The conjugate of claim 96, wherein the functional group is indirectly linked to the multiply amphipathic polymer through an inert linking moiety.

99. (Original) The conjugate of claim 96, wherein the affinity molecule is selected from the group consisting of an antigen, hapten, antibody, antibody fragment, hormone, hormone binding protein, enzyme, enzyme inhibitor, nucleic acid, nucleotide, oligonucleotide, polynucleotide, receptor agonist, and receptor antagonist.

100. (Original) The conjugate of claim 96, wherein the first and second members of the binding pair are selected from the group consisting of: antigen and antibody; antigen and antibody fragment; hapten and antibody; hapten and antibody fragment; biotin and avidin; biotin and streptavidin; hormone and hormone binding protein; receptor agonist and receptor; receptor antagonist and receptor; IgG and protein A; lectin and carbohydrate; enzyme and enzyme cofactor; enzyme and enzyme inhibitor; and complementary oligonucleotides.

101. (Original) The conjugate of claim 96, wherein the second member of the binding pair is associated with the first member of the binding pair through a covalent bond.

102. (Original) The conjugate of claim 96, wherein the second member of the binding pair is associated with the first member of the binding pair through a noncovalent bond.

103. (Original) The conjugate of claim 96, wherein the functional group is selected from amino, cyano, carboxyl, ester, and halocarbonyl groups.

104. (Original) The conjugate of claim 103, wherein the functional group is an amino group.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

105. (Original) The water-dispersible nanoparticle of claim 96, wherein the inner core is comprised of a semiconductive material.

106. (Original) The conjugate of claim 105, wherein the semiconductive material is inorganic.

107. (Original) The conjugate of claim 106, wherein the semiconductive material is crystalline.

108. (Original) The conjugate of claim 105, wherein the water-insoluble organic coating is comprised of trioctylphosphine oxide, trioctylphosphine, tributylphosphine, or a mixture thereof.

109. The conjugate of claim 105, further including a shell layer between the core and the water-insoluble organic coating.

110. (Original) The conjugate of claim 109, wherein the shell layer is comprised of a semiconductive material having a band gap energy greater than that of the inner core.

111. (Original) The conjugate of claim 96, wherein the inner core is comprised of a metallic material.

112. (Original) The conjugate of claim 111, wherein the water-insoluble organic coating has an affinity for the metallic material.

113. (Original) The conjugate of claim 107, wherein the water-insoluble organic coating is comprised of a hydrophobic surfactant.

114. (Previously presented) The conjugate of claim 113, wherein the hydrophobic surfactant is selected from the group consisting of octanethiol, dodecanethiol, dodecylamine, tetraoctylammonium bromide, and mixtures thereof.

115. (Original) The conjugate of claim 96, wherein the multiply amphipathic polymer is linear or branched.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

116. (Original) The conjugate of claim 115, wherein the multiply amphipathic polymer is branched.

117. (Original) The conjugate of claim 116, wherein the multiply amphipathic polymer is hyperbranched or dendritic.

118. (Original) The conjugate of claim 96, wherein the hydrophobic regions of the multiply amphipathic polymer are each comprised of at least one non-ionizable, nonpolar monomer unit.

119. (Original) The conjugate of claim 96, wherein each of the hydrophobic regions comprise at least one monomer unit selected from the group consisting of ethylene, propylene, alkyl (C<sub>4</sub>-C<sub>12</sub>)-substituted ethyleneimine, an alkyl acrylate, a methacrylate, a phenyl acrylate, an alkyl acrylamide, a styrene, a hydrophobically derivatized styrene, a vinyl ether, a vinyl ester, a vinyl halide, and a combination thereof.

120. (Original) The conjugate of claim 119, wherein the hydrophobic regions comprise at least one monomer unit selected from an alkyl acrylate, an alkyl methacrylate, an alkyl acrylamide, and a mixture thereof.

121. (Original) The conjugate of claim 96, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit containing an ionizable or polar moiety.

122. (Original) The conjugate of claim 121, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

123. (Original) The conjugate of claim 122, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and amine substituents.

124. (Original) The conjugate of claim 118, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable or polar moiety.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

125. (Original) The conjugate of claim 124, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

126. (Original) The conjugate of claim 125, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and an amine substituent.

127. (Original) The conjugate of claim 96, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit selected from the group consisting of a water-soluble ethylenically unsaturated C<sub>3</sub>-C<sub>6</sub> carboxylic acid, an allylamine, an inorganic acid addition salt of an allylamine, a di-C<sub>1</sub>-C<sub>3</sub>-alkylamino-C<sub>2</sub>-C<sub>6</sub>-alkyl acrylate, a methacrylate, an olefinically unsaturated nitrile, a diolefinically unsaturated monomer, N-vinyl pyrrolidone, N-vinyl formamide, an acrylamide, a lower alkyl-substituted acrylamide, a lower alkoxy-substituted acrylamide, N-vinylimidazole, N-vinylimidazoline, a styrene sulfonic acid and an alkylene oxide.

128. (Original) The conjugate of claim 127, wherein the hydrophilic regions each comprise at least one monomer unit selected from the group consisting of acrylic acid, methacrylic acid, styrene sulfonic acid, acrylamide and methacrylamide.

129. (Original) The conjugate of claim 96, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise a vinyl monomer substituted with at least one hydrophilic moiety selected from the group consisting of a carboxylate, a thiocarboxylate, an amide, an imide, a hydrazine, a sulfonate, a sulfoxide, a sulfone, a sulfite, a phosphate, a phosphonate, a phosphonium, an alcohol, a thiol, a nitrate, an amine, an ammonium, and an alkyl ammonium group  $-\text{[NHR}^1\text{R}^2\text{]}^+$ , wherein R<sup>1</sup> and R<sup>2</sup> are alkyl substituents.

130. (Original) The conjugate of claim 129, wherein the hydrophilic moiety is directly bound to a carbon atom in the backbone of the multiply amphipathic polymer.

131. (Original) The conjugate of claim 129, wherein the hydrophilic moiety is bound to a carbon atom in the polymer backbone of the multiply amphipathic polymer through a

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

linkage selected from the group consisting of alkylene, alkenylene, heteroalkylene, heteroalkenylene, arylene, heteroarylene, alkarylene, and aralkylene.

132. (Original) The conjugate of claim 96, wherein the multiply amphipathic polymer is a copolymer of a hydrophilic monomer selected from the group consisting of acrylic acid, methacrylic acid and combinations thereof, with at least one hydrophobic alkyl (C<sub>6</sub>-C<sub>12</sub>) acrylamide monomer.

133. (Original) The conjugate of claim 132, wherein the multiply amphipathic polymer is poly(acrylic acid-co-octylacrylamide).

134. (Original) The conjugate of claim 96, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 500 to 50,000.

135. (Original) The conjugate of claim 134, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 10,000.

136. (Original) The conjugate of claim 135, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 5000.

137. (Original) The conjugate of claim 133, wherein the poly(acrylic acid-co-octylacrylamide) has a molecular weight in the range of approximately 1000 to 5000.

138. (Original) The conjugate of claim 96, wherein the hydrophobic regions represent in the range of approximately 25 wt. % to 90 wt. % of the multiply amphipathic polymer.

139. (Original) The conjugate of claim 96, wherein the multiply amphipathic polymer is a polypeptide in which the hydrophobic regions are comprised of at least one hydrophobic amino acid and the hydrophilic regions are comprised of at least one hydrophilic amino acid.

140. (Original) The conjugate of claim 96, wherein the multiply amphipathic polymer is crosslinked.



Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

141. (Original) A composition comprising a plurality of nanoparticle conjugates each comprising:

(a) a water-dispersible nanoparticle and an affinity molecule that serves as a first member of a binding pair, wherein the nanoparticle comprises (i) an inner core comprised of a semiconductive or metallic material, (ii) a water-insoluble organic coating provided thereon, (iii) an outer layer of a multiply amphipathic polymer having at least two hydrophobic regions and at least two hydrophilic regions, and (iv) a functional group covalently linked to the polymer, and further wherein the affinity molecule is bound to the functional group; and

(b) a second member of the binding pair associated with the first member through either covalent or noncovalent interaction.

142. (Original) The composition of claim 141, wherein the inner cores of the nanoparticles are members of a monodisperse particle population.

143. (Original) The composition of claim 142, wherein the monodisperse particle population is characterized in that when irradiated the population emits light in a bandwidth in the range of approximately 20 nm to 60 nm full width at half maximum (FWHM).

144. (Original) The composition of claim 143, wherein the monodisperse particle population is characterized in that when irradiated the population emits light in a bandwidth in the range of approximately 20 nm to 40 nm full width at half maximum (FWHM).

145. (Original) The composition of claim 142, wherein the monodisperse particle population is characterized in that it exhibits no more than about a 10% rms deviation in the diameter of the inner core.

146. (Original) The composition of claim 145, wherein the monodisperse particle population is characterized in that it exhibits no more than about a 5% rms deviation in the diameter of the inner core.

147. (Original) The composition of claim 141, wherein the affinity molecule is selected from the group consisting of an antigen, hapten, antibody, antibody fragment, hormone,

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

hormone binding protein, enzyme, enzyme inhibitor, nucleic acid, nucleotide, oligonucleotide, polynucleotide, receptor agonist, and receptor antagonist.

148. (Original) The composition of claim 141, wherein the first and second members of the binding pair are selected from the group consisting of: antigen and antibody; antigen and antibody fragment; hapten and antibody; hapten and antibody fragment; biotin and avidin; biotin and streptavidin; hormone and hormone binding protein; receptor agonist and receptor; receptor antagonist and receptor; IgG and protein A; lectin and carbohydrate; enzyme and enzyme cofactor; enzyme and enzyme inhibitor; and complementary oligonucleotides.

149. (Original) The composition of claim 141, wherein the second member of the binding pair is associated with the first member of the binding pair through a covalent bond.

150. (Original) The composition of claim 141, wherein the second member of the binding pair is associated with the first member of the binding pair through a noncovalent bond.

151. (Original) The composition of claim 141, wherein the affinity molecule is covalently linked to the multiply amphipathic polymer through an inert linking moiety.

152. (Original) The composition of claim 141, wherein the functional group is selected from amino, cyano, carboxyl, ester, and halocarbonyl groups.

153. (Original) The composition of claim 152, wherein the functional group is an amino group.

154. (Original) The composition of claim 141, wherein the inner core is comprised of a semiconductive material.

155. (Original) The composition of claim 154, wherein the semiconductive material is inorganic.

156. (Original) The composition of claim 155, wherein the semiconductive material is crystalline.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

157. (Original) The composition of claim 154, wherein the water-insoluble organic coating is comprised of trioctylphosphine oxide, trioctylphosphine, tributylphosphine, or a mixture thereof.

158. (Original) The composition of claim 154, further including a shell layer between the core and the water-insoluble organic coating.

159. (Original) The composition of claim 158, wherein the shell layer is comprised of a semiconductive material having a band gap energy greater than that of the inner core.

160. (Original) The composition of claim 141, wherein the inner core is comprised of a metallic material.

161. (Original) The composition of claim 160, wherein the water-insoluble organic coating has an affinity for the metallic material.

162. (Original) The composition of claim 161, wherein the water-insoluble organic coating is comprised of a hydrophobic surfactant.

163. (Original) The composition of claim 162, wherein the hydrophobic surfactant is selected from the group consisting of octanethiol, dodecanethiol, dodecylamine, tetraoctylammonium bromide, and mixtures thereof.

164. (Original) The composition of claim 141, wherein the multiply amphipathic polymer is linear or branched.

165. (Previously presented) The composition of claim 164, wherein the polymer is branched.

166. (Original) The composition of claim 165, wherein the polymer is hyperbranched or dendritic.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

167. (Original) The composition of claim 141, wherein the hydrophobic regions of the multiply amphipathic polymer are each comprised of at least one non-ionizable, nonpolar monomer unit.

168. (Original) The composition of claim 141, wherein each of the hydrophobic regions comprise at least one monomer unit selected from the group consisting of ethylene, propylene, alkyl (C<sub>4</sub>-C<sub>12</sub>)-substituted ethyleneimine, an alkyl acrylate, a methacrylate, a phenyl acrylate, an alkyl acrylamide, a styrene, a hydrophobically derivatized styrene, a vinyl ether, a vinyl ester, a vinyl halide, and a combination thereof.

169. (Original) The composition of claim 168, wherein the hydrophobic regions comprise at least one monomer unit selected from an alkyl acrylate, an alkyl methacrylate, an alkyl acrylamide, and a mixture thereof.

170. (Original) The composition of claim 141, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit containing an ionizable or polar moiety.

171. (Original) The composition of claim 170, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

172. (Original) The composition of claim 171, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and amine substituents.

173. (Original) The composition of claim 167, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable or polar moiety.

174. (Original) The composition of claim 173, wherein the hydrophilic regions each comprise at least one monomer unit containing an ionizable moiety.

175. (Original) The composition of claim 174, wherein the ionizable moiety is selected from the group consisting of carboxylic acid, sulfonic acid, phosphonic acid, and an amine substituent.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

176. (Original) The composition of claim 141, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise at least one monomer unit selected from the group consisting of a water-soluble ethylenically unsaturated C<sub>3</sub>-C<sub>6</sub> carboxylic acid, an allylamine, an inorganic acid addition salt of an allylamine, a di-C<sub>1</sub>-C<sub>3</sub>-alkylamino-C<sub>2</sub>-C<sub>6</sub>-alkyl acrylate, a methacrylate, an olefinically unsaturated nitrile, a diolefinically unsaturated monomer, N-vinyl pyrrolidone, N-vinyl formamide, an acrylamide, a lower alkyl-substituted acrylamide, a lower alkoxy-substituted acrylamide, N-vinylimidazole, N-vinylimidazoline, a styrene sulfonic acid and an alkylene oxide.

177. (Original) The composition of claim 176, wherein the hydrophilic regions each comprise at least one monomer unit selected from the group consisting of acrylic acid, methacrylic acid, styrene sulfonic acid, acrylamide and methacrylamide.

178. (Original) The composition of claim 141, wherein the hydrophilic regions of the multiply amphipathic polymer each comprise a vinyl monomer substituted with at least one hydrophilic moiety selected from the group consisting of a carboxylate, a thiocarboxylate, an amide, an imide, a hydrazine, a sulfonate, a sulfoxide, a sulfone, a sulfite, a phosphate, a phosphonate, a phosphonium, an alcohol, a thiol, a nitrate, an amine, an ammonium, and an alkyl ammonium group  $-\text{[NHR}^1\text{R}^2\text{]}^+$ , wherein R<sup>1</sup> and R<sup>2</sup> are alkyl substituents.

179. (Original) The composition of claim 178, wherein the hydrophilic moiety is directly bound to a carbon atom in the backbone of the multiply amphipathic polymer.

180. (Original) The composition of claim 178, wherein the hydrophilic moiety is bound to a carbon atom in the backbone of the multiply amphipathic polymer through a linkage selected from the group consisting of alkylene, alkenylene, heteroalkylene, heteroalkenylene, arylene, heteroarylene, alkarylene, and aralkylene.

181. (Original) The composition of claim 141, wherein the multiply amphipathic polymer is a copolymer of a hydrophilic monomer selected from the group consisting of acrylic acid, methacrylic acid and combinations thereof, with at least one hydrophobic alkyl (C<sub>6</sub>-C<sub>12</sub>) acrylamide monomer.

Attorney Docket No. 130924.6211  
Serial No. 10/716,971  
Inventor: Adams  
Paper dated: March 6, 2006

182. (Original) The composition of claim 181, wherein the multiply amphipathic polymer is poly(acrylic acid-co-octylacrylamide).

183. (Original) The composition of claim 141, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 500 to 50,000.

184. (Original) The composition of claim 183, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 10,000.

185. (Original) The composition of claim 184, wherein the multiply amphipathic polymer has a molecular weight in the range of approximately 1000 to 5000.

186. (Original) The composition of claim 182, wherein the poly(acrylic acid-co-octylacrylamide) has a molecular weight in the range of approximately 1000 to 5000.

187. (Original) The composition of claim 141, wherein the hydrophobic regions represent in the range of approximately 25 wt. % to 90 wt. % of the multiply amphipathic polymer.

188. (Original) The composition of claim 141, wherein the multiply amphipathic polymer is a polypeptide in which the hydrophobic regions are comprised of at least one hydrophobic amino acid and the hydrophilic regions are comprised of at least one hydrophilic amino acid.

189. (Original) The composition of claim 141, wherein the multiply amphipathic polymer is crosslinked.